УДК 57.083.3

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NUCLEAR MAGNETIC RESONANCE-BASED ASSAYS IN IMMUNODIAGNOSTICS*

Keywords: NMR-based assay, magnetic nanoparticles, prostate-specific antigen, tetanus toxoid.

Currently, immunodiagnostics requires high sensitivity, reliability and reproducibility assays. One approach of increasing sensitivity is to use nuclear magnetic resonance relaxometry and magnetic nanoparticles [1]. The essence of assays based on this method is to register the T2 proton relaxation time changes depending on the presence of magnetic nanoparticles (MNP) in the medium [2].

Two approaches to the development of analytical test systems using nuclear magnetic resonance were developed. Prostate-specific antigen (PSA) and IgG specific to tetanus toxoid (Anti-TT IgG) were acting as model analyte.

Applied to PSA detection, assay principle is as follows. A nitrocellulose membrane coated with capture antibodies and afterward incubated with a sample and magnetic nanoparticles functionalized by recognition molecule. Test strips were placed in a portable NMR relaxometer. Magnetic nanoparticles attached to test strip decrease the T2 relaxation time of the water protons inside the pores of the membrane. Thus, T2 is inversely proportional to the concentration of the antigen (PSA) in the sample.

The approach of the assay for anti-TT IgG is that ELISA plates are coated with capture element, then analyte is added and detected by the conjugate of magnetic nanoparticles with recognition molecule. Then magnetic nanoparticles were displaced from the wells surface into medium by addition of the elution solution (0.1 M sodium hydroxide).

Detached magnetic nanoparticles decrease the transverse relaxation times (T2) of protons from surrounding solution and T2 is inversely proportional to the concentration of the anti-TT IgG in the sample.

Calibration plots were constructed under optimal experimental conditions for each test system (figure).

Thus, during the research, two different approaches for NMR-based assays were developed. Analytical performance of designed (table) method is acceptable for immunodiagnostics test-systems [3].

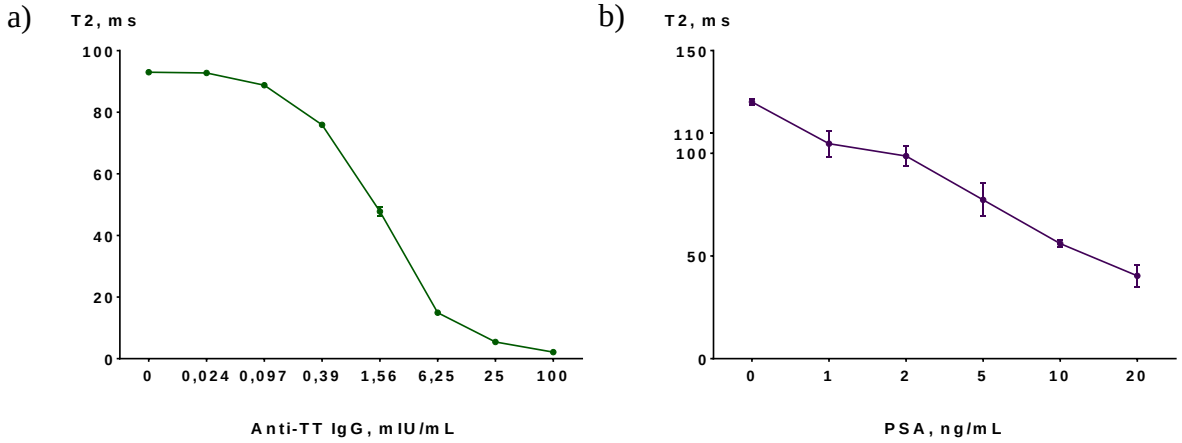

Figure. Calibration plot for a) IgG specific to tetanus toxoid (Anti-TT IgG) detection
b) Prostate-specific antigen (PSA) detection

Table

Immunoassay performance

Characteristic value	Method	
	Test for PSA	Test for anti-TT antibodies
Analytical sensitivity: LOD	0.44 ng/ml	0.08 mIU/ml
Reproducibility: CV,%	7.8	7

References

1. *Alcantara D., Lopez S., García-Martin M. L., Pozo D.* // Nanomedicine: Nanotechnology, Biology, and Medicine. 2016. Vol. 12. № 5. P. 1253–1262.
2. *Denmark D. J., Bustos-Perez X., Swain A. et al.* // Journal of Electronic Materials. 2019. Vol. 48. P. 4749–4761.
3. *Khramtsov P., Kropaneva M., Bochkova M. et al.* // Microchimica Acta. 2019. Vol. 186. № 12. P. 1–7.

** This work was supported by Russian Science Foundation (project № 17-15-01116).*